RESULTS OF A 9-MONTH FEEDING STUDY WITH OCDD AND OCDF IN RATS

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INTRODUCTION

OCDD and OCDF are ubiquitous environmental pollutants which are released from various sources, as e.g. municipal waste incinerators. Levels of about 600 ppb OCDD and 40 ppb OCDF were detected in fly ash from such plants [1]. Human milk, originating from industrialized countries, was found to contain up to 1300 ppt OCDD (on fat basis) with an average of about 270 ppt. Generally, much less OCDF is found in breast milk (3-4 ppt, highest values around 70 ppt, on fat basis) [2].

Both isomers are presently assigned a toxic equivalency factor (TEF) of 0.001, based on enzyme induction data (available only for OCDD). Because of their toxicokinetic properties and the high concentration of, especially OCDD, in human adipose tissue a thorough evaluation of the toxic potential of these compounds seemed to be necessary. For OCDD a whole body half-life of 3 to 5 months in rats was reported by Birnbaum & Couture [3] and of 8 months by Wacker [4]. No kinetic data are available on OCDF. Gastrointestinal absorption of OCDD is poor and tends to decrease with increasing dose [3]. Thus, an administration of the compounds in a longterm study appeared to be a prerequisite to attain sufficiently high tissue levels for the production of toxic effects, though it was clear beforehand that even within an experimental period of 9 months no steady state tissue levels would be reached.

In a recent study Couture et al. [5] administered 50 µg OCDD/kg to rats for 13 weeks by gavage (5 times/wk). This treatment resulted in 40-fold elevated EROD-activities over controls and a doubling of cytochrome P-450 content in liver. Except for some lipid droplets in the liver, they observed no other toxic effects as usually seen with the toxic PCDD/F. Based on these findings the authors assumed that the TEF of OCDD would be between 0.01 and 0.001.

In order to extend the data base for OCDD and the isosteric furan the present study was undertaken.

EXPERIMENTAL APPROACH

OCDD and OCDF were purchased from amchro Chromatographie GmbH (Sulzbach, Germany), 2378-TCDD originated from Dow Chemical (Midland, Mi, USA). Purity analysis by HRGC/MS exhibited no detectable amounts of 2378-TCDD (det. limits: 0.6 ppm (OCDD), 1.6 ppm (OCDF)), but traces of 23478-PeCDF in the OCDD (2.5 ppm) as well as 1234678-HpCDD (170 ppm in OCDD) and 1234678-HpCDF (175 ppm in OCDF). The total concentrations of toxic congeners, using detection limit values for those congeners not detectable, were below 76 ppm TCDD-equivalents in the OCDD and 95 ppm in the OCDF (Swiss TEFs [7]). These levels had no toxicological importance.

Groups of 6 female rats (Iva: SIV 50 (SD), starting weight of about 135 g) were maintained for 13, 26 or 39 weeks on diets (Nafag 890) containing 0, 80 and 800 ppb of OCDD, 80 and 800 ppb of OCDF or 1 ppb of 2378-TCDD (total 18 groups). The 2378-TCDD was spiked with a small amount of the tritium-labeled compound. Food consumption (per group) and body weights were recorded weekly. Terminal body and organ weights were determined and selected organs subjected to histopathological examination. EROD-activity was determined in animals of each dose group. In liver and adipose tissue residues of OCDD, OCDF and 2378-TCDD were measured by GC-MS or GC-ECD, or by radioactivity determination.

RESULTS AND DISCUSSION

Feed consumption per dose group was not significantly influenced by the contaminants. Weight development was slighly depressed in the high OCDD/OCDF dose groups and reached a level of significance ($p<0.05$) in some groups (see Fig. 1). Thymus weights were significantly lower than in control animals after 13 weeks in rats receiving the 800 ppb OCDD and OCDF diet as well as 2378-TCDD. After 26 weeks thymus weights of all treated groups were significantly lower than those of the controls. However, due to the age-dependent involution of control thymus the decrease after 36 weeks remained significant only in the 800 ppb OCDD group (Fig. 2). Other organ weights were much less affected and changes were significant only for liver in some of the high dose groups and the 2378-TCDD positive control.

EROD-activity was significantly elevated over controls already in both low dose groups. Both, OCDD and OCDF, at the high dose level, as well as 2378-TCDD produced a 20-50 fold elevation of EROD-activity over controls (Fig. 3). Interestingly, induction did not increase further or rather decreased after 26 or 39 weeks. This gives some indications that enzyme induction does not respond well to the accumulation of the compounds in liver tissue.

Liver and adipose tissue levels of OCDD, OCDF and 2378-TCDD, determined after 13, 26 and 39 weeks are given in Table 1 (data on OCDD/OCDF at week 39 will be available at the Symposium). Retention of 2378-TCDD in the liver (expressed as percent of the dose) was higher than that of OCDD and OCDF at week 13, most likely due to their poor absorption from the gastrointestinal tract. Both compounds had accumulated more in liver than in adipose tissue, as it becomes evident from the liver to adipose tissue concentration

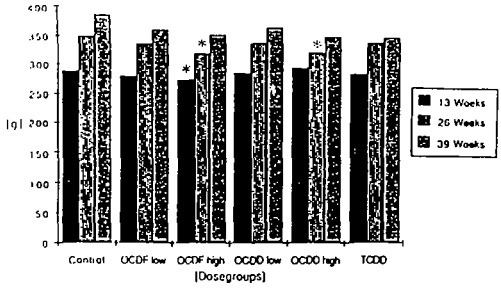

Figure 1: Mean body weight of rats maintained on diets containing OCDD or OCDF (80 and 800 ppb) or 2378-TCDD (1 ppb).
**p 0.05 against controls (all alive rats used for calculation).*

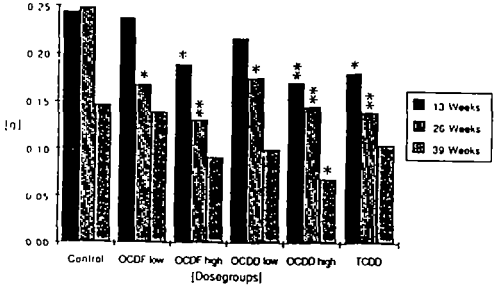

Figure 2: Mean absolute thymus weights of rats kept on diets containing OCDD or OCDF (80 and 800 ppb), or 2378-TCDD (1 ppb).
**p 0.05, **p 0.01, versus controls.*

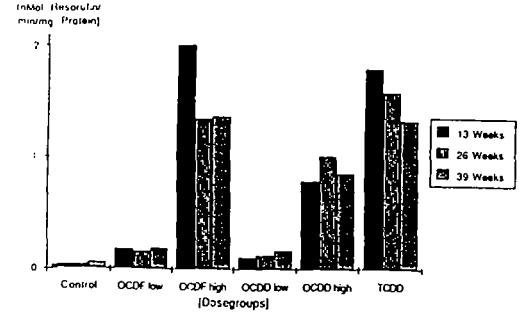

Figure 3: 7-Ethoxyresorufin o-de-ethylase (EROD)-activity in hepatic tissue of rats maintained on diets containing OCDD or OCDF (80 and 800 ppb) or 2378-TCDD (1 ppb).

Table 1: Liver and adipose tissue concentration of OCDD, OCDF and 2378-TCDD in rats fed diets containing OCDD or OCDF (80 and 800 ppb) or 1 ppb 2378-TCDD.

	Concentration in liver [ng/g]			% of total dose in whole liver			Concentration in fat [ng/g]			Concentration ratio liver/fat		
Time interval	13 W	26 W	39W	13 W	26 W	39W	13 W	26 W	39W	13 W	26 W	39W
OCDF low	560	720		3.7	2.6		9.1	18.5		62	39	
OCDF high	7260	8640		5.2	4.0		128.5	135.2		56	64	
OCDD low	470	590		3.3	2.3		22.5	30.6		21	19	
OCDD high	5230	7800		3.6	3.3		130.4	201.2		40	39	
2,3,7,8-TCDD	12.5	11.2	12.3	7.5	3.9	2.8	4.9	4.2	4.2	2.5	2.6	2.9

ratios. This is in accordance with earlier observations on OCDD and OCDF as well as other congeners [4,6]. Liver and adipose tissue levels of 2378-TCDD were rather similar after 13, 26 and 39 weeks, indicating that steady state conditions were reached already within the first time interval. This was not true for the two octachloro-compounds. Histopathological examination of the organs has not yet been completed, however, reports will be available for Dioxin '90. Under consideration of the available data, both compounds would have a toxic potential between 1/100 and 1/1000 that of 2378-TCDD.

CONCLUSIONS

- Both, OCDD and OCDF produced "dioxin-like" effect in rats at concentrations of 80 and 800 ppb in feed.
- The toxicity of both compounds is similar.
- The TEF for both compounds is between 0.01 and 0.001. More exact estimates will be possible after inclusion of histopathology evaluations.
- Since there are major differences in toxicokinetics between 2378-TCDD and OCDD/OCDF this must be taken into account, when their toxic potential is estimated.

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